An equation for the calculation of retention indices in temperature-programmed gas chromatography with allowance for the nonlinear variation of the retention parameters of *n*-alkanes

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In view of the nonlinear variation of the temperature increments of *n*-alkanes found previously, the accuracy of the calculations of the retention indices (I_{pr}) of substances in temperature-programmed capillary gas chromatography carried out in terms of six known equations was verified. A new four-parameter equation was proposed, and a general method for the calculation of its coefficients, suitable for all stationary phases, based on the adjusted retention times of *n*-alkanes was suggested. The coefficients of the equation for 12 temperature variation programs were determined. Using the homologous series of methyl esters of fatty acids as an example, it was shown that the proposed equation ensures the minimum error of determination of I_{pr} under various conditions. The equation also makes it possible to carry out interpolation and extrapolation calculations. The coefficients of the equation are found using the least-squares method based on data for any 4—5 reference *n*-alkanes.

Key words: capillary gas chromatography, linear temperature programming, nonlinear equation for calculation of retention indices; *n*-alkanes; methyl esters of fatty acids.

The retention indices (I_{pr}) for temperature-programmed capillary gas chromatography (TPGC) are calculated most frequently using the van der Doole and Kratz equation,[1] in which the temperatures of the emergence or retention times of the compound analyzed and those of reference *n*-alkanes, between which it comes out, serve as the parameters. This equation is based on the assumption that the emergence temperatures (retention times) of *n*-alkanes are linearly dependent on the number of C atoms in their molecules. However, in actual practice, this linearity does not hold.[2,3] Other formulas for calculation of the indices under conditions of TPGC, based on the linearity principle, are also known,[4,5] but they have gained no acceptance. We showed[6] that one of the main reasons for the nonlinear variation of the retention parameters of *n*-alkanes as a function of the number of C atoms in their molecules is the inequality of the temperature increments for various homologs, *i.e.*, the variation of the retention time as a function of the temperature of the analysis ($d\ln t_n'/dT$). The deviation from a linear dependence increases with an increase in the rate of the temperature variation and the molecular weight of the *n*-alkane.

Judging from the published data dealing with determination of the retention indices for TPGC, most of these studies are carried out along two lines:

— development of methods for prediction of the retention parameters of substances based on the data on their retention under isothermal conditions;[4,7—11]

— development of mathematical models, other than the van der Doole and Kratz equation,[3,12,13] taking into account the nonlinear variation of the retention parameters of *n*-alkanes.

Our study belongs to the latter group and was undertaken because the known mathematical models[3,12,13] suffer from a number of substantial drawbacks, because they do not take into account the inequality of the temperature increments for the reference *n*-alkanes.[6] In particular, one of the equations[12] suggested previously does not describe the nonlinear variation of the retention parameters of *n*-alkanes with sufficient accuracy; therefore, the standard deviations (s) of the retention indices obtained from this equation can be as high as ten or more index units (iu).[13]

A more accurate regression equation has also been proposed[13]

$$I_{pr} = a_0 + a_1 \ln Y + a_2 Y^B,$$

where a_0, a_1, a_2, and B are constants; Y is the retention parameter (V_g or t'). However, since the dependence of exponent B on the conditions of analysis and on the choice of reference compounds is complex, the calculations of I_{pr} are quite labor-consuming. To use the method of cubic interpolation,[3] special programs need to be composed for calculation of the coefficients of the equation.

The purpose of this work was to verify the accuracy of determination of the I_{pr} indices in terms of the

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equations proposed previously, taking into account the inequality of the temperature increments of reference *n*-alkanes, which we found earlier,[6] and to find a new dependence that would enable sufficiently accurate calculation of the retention parameters of the components of complex mixtures, analyzed by TPGC, and would also be suitable outside the scale of reference *n*-alkanes, *i.e.*, when some of the homologs are missing.

Experimental

Chromatographically pure *n*-alkanes, C_nH_{2n+2} (n = 5 to 14), and methyl esters of fatty acids, R_mCOOMe (m = 1 to 9), were used as the objects of investigation. The retention parameters of the substances were determined using a Micromat-412 chromatograph (Nordion, Finland) with a flame ionization detector. A quartz capillary column (25 m × 0.32 mm) packed with a standard SE-30 phase (thickness of the layer of d_f = 1.0 μ) was used. The analyses were carried out under conditions of linear programming of the temperature of the column; the initial temperatures T_0 were 120, 140, 160, and 180 °C and the rates of temperature variation r were 2, 6, and 8 °C min^{-1}. The temperatures of the evaporation chamber and detector were equal to 225 and 250 °C, respectively. The pressure of the carrier gas (He) at the inlet of the column was 0.5 bar; the ratio of flow splitting before the inlet of the column was 1 : 100. Samples of 0.1 μL of 0.2% solutions of methyl esters or *n*-alkanes in pentane were analyzed.

Each retention index was measured in three to seven entries. The error of calculation with respect to the experimental values of the indices amounted to ±1 iu. The errors of the calculation of the indices in terms of the various equations were estimated from the standard deviations (s) of the experimental results from the calculated values.

Results and Discussion

The retention indices in TPGC depend on the initial temperature in the program (T_0), the rate of temperature variation (r), the temperature increments of the reference *n*-alkanes and substances under analysis (dI/dT), and the retention time (t) of the carrier gas in the column.

The retention indices of the components of a complex mixture analyzed by TPGC are usually calculated from the equation

$$I_{pr} = 100n + (T_x - T_n)/(T_{n+l} - T_n)\cdot 100, \qquad (1)$$

where I_{pr} is the retention index of compound x under conditions of TPGC; T_x, T_n, and T_{n+1} are the temperatures of the emergence (or retention times t_x, t_n, and t_{n+1}) of compound x and of reference *n*-alkanes containing n and (n + 1) carbon atoms in the molecule, respectively; in addition, $T_n < T_x < T_{n+1}$. Equation (1) suggests that the retention parameters of all homologous *n*-alkanes, on whose scale the sorption characteristics of the compounds analyzed are located, are known. In the case of extrapolation calculation, *i.e.*, when $T_x > T_{n+1}$ or $T_x < T_n$, this expression leads to large errors in the determination of I_{pr}. Inaccurate values of the retention indices of the compounds analyzed are also obtained if the calculation is based on two *n*-alkanes that differ by more than one C atom. To make it clearer, let us present the calculation of the retention indices I_{pr} of substances from Eq. (1) as a scheme (Fig. 1). The temperatures of the emergence of *n*-alkanes containing n, (n +1), and (n + 2) carbon atoms are denoted by T_n, T_{n+1}, and T_{n+2}, and the retention indices corresponding to them are denoted by I_n = 100n, I_{n+1} = 100(n + 1), and I_{n+2} = 100(n + 2). According to Eq. (1), the straight line drawn through points *1* and *2* for *n*-alkanes with n and (n + 1) carbon atoms, is part of the scale of *n*-alkanes corresponding to compounds whose emergence temperatures (or retention times) under conditions of TPGC satisfy the inequality $T_n < T_x < T_{n+1}$ ($t_n < t_x < t_{n+1}$). The retention index of substance x with the temperature of emergence T_x is represented in the diagram by the intercept $0—I_x'$. It can be seen from Fig. 1 that using Eq. (1), we obtain the value I_x' instead of the retention index I_x. If the calculation of the retention index of compound x in terms of Eq. (1) is based on another pair of *n*-alkanes, for example on those containing n and (n + 2) carbon atoms, this will result in a different value of I_x''. The smaller n and the larger r, the more pronounced the nonlinearity of the $I = f(T)$ dependence and the larger the difference between the I_x, I_x', and I_x'' values.

Thus, to perform an accurate calculation of the retention indices of the components of a complex mixture analyzed by TPGC, it is necessary to find an equation that would describe the real dependence of the retention indices of *n*-alkanes on the temperatures of their emergence (or retention times) and would take into account the different temperature increments of homologous *n*-alkanes and the retention time of a nonsorbed gas in the column.

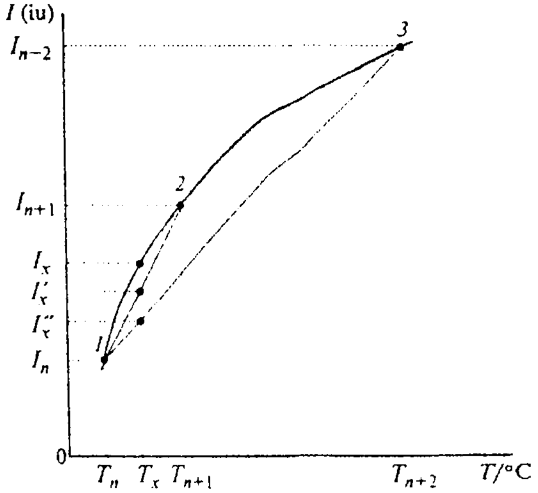

Fig. 1. Scheme of calculation of retention indices of compounds analyzed by TPGC in terms of Eq. (1) (for *1*—*3*, see text).

Choice of the form for the equation characterizing retention of *n*-alkanes analyzed under temperature-programmed conditions. Various nonlinear equations, describing the gas-chromatpographic behavior of homologous organic compounds analyzed under isothermal conditions, have been reported[14–16]:

$$Z = a + bq + cq^2 + dq^3, \tag{2}$$

$$Z = a + b/q + cq, \tag{3}$$

$$Z = a + bq + c\ln q/q + d/(q-2)^2 + 0.1, \tag{4}$$

$$Z = a + bq + c\ln q + d\ln(\ln q), \tag{5}$$

where Z is the retention index of the compound analyzed, or the logarithm of its specific retention volume, or the logarithm of the adjusted retention time; q is the number of the homolog (n for *n*-alkanes or m for other homologous series).

Equation (2) has also been used in relation to temperature-programmed conditions ($Z = I_{pr}$, q is the adjusted retention time of a homolog).[14] Expressions (2)—(5) describe the variation of the retention parameters of compounds in homologous series. In view of the fact that isothermal conditions of analysis can be regarded as linear temperature variation at a rate of $r = 0$ °C min^{-1}, we verified the applicability of Eqs. (4) and (5) to the description of the behavior of compounds in TPGC as $I = f(t')$. In addition, we analyzed Eq. (2) as well as relations (6)—(9), which we obtained from Eqs. (2)—(5):

$$I_{pr} = a + b/t' + ct' + d(t')^2, \tag{6}$$

$$I_{pr} = a + bt' + c\ln t'/t' + d/(t')^2, \tag{7}$$

$$I_{pr} = a + bt' + \ln t' + d(t')^2, \tag{8}$$

$$I_{pr} = a + bt' + c\ln t' + d/t', \tag{9}$$

where $I_{pr} = 100n$ is the retention index of an *n*-alkane under conditions of TPGC; $t' = (t - \sigma)$ is the adjusted retention time of the *n*-alkane, t is the retention time of the *n*-alkane, σ is the residence time of a non-sorbed gas in the column at the temperature of emergence of the *n*-alkane.[4,17]

In order to compare the potentialities of Eq. (2) and Eqs. (4)—(9) for calculation of the retention indices of *n*-alkanes under conditions of TPGC, we determined the standard deviations (s) of the values found using relations (2) and (4)—(9) from the indices accepted for *n*-alkanes, which are equal to 100n. The results showed that in the case of TPGC, the s values depend on the rate of temperature variation r and on the initial temperature of the analysis T_0 (Table 1). An increase in r and T_0 leads to a decrease in s in the case of Eqs. (2), (4), (6), and (7); however, for relations (5), (8), and (9), this dependence is not observed. Equation (9) ensures the most accurate determination of the indices I_{pr} from the adjusted retention times. The errors of calculation of the retention parameters of *n*-alkanes for various programs of temperature variation in terms of Eq. (9) are 1—2 orders of magnitude smaller than those for relations (2), (4), and (6)—(8) and 1.5—2 times smaller than the errors of the calculation in terms of Eq. (5). Expression (9) describes the $I_{pr} = f(t')$ function that varies continuously and smoothly following an increase in the molecular weight of the *n*-alkanes; the coefficients of this equation can be found by the least-squares method.

Table 1. Standard deviations (s) of the values of retention indices of the homologous series of C_5—C_{11} *n*-alkanes calculated from Eqs. (2) and (4)—(9) for various temperatue variation programs in TPGC

Equation	T_0 = 140,	T_0 = 160		T_0 = 180	
	r = 2	r = 6	r = 8	r = 2	r = 8
(2)	92.2	69.3	64.6	74.9	58.0
(6)	51.4	39.6	36.9	42.5	34.9
(4)	39.4	27.0	24.1	29.9	22.7
(7)	35.7	24.3	21.5	27.2	21.5
(8)	4.3	5.0	5.4	5.4	2.5
(5)	1.2	1.2	1.6	1.2	1.6
(9)	0.8	0.6	1.2	1.2	0.6

Note. Here and in Table 2, T_0/°C is the initial temperature in the program, r/°C min^{-1} is the rate of variation of the temperature of the column.

Thus, the data that we obtained indicate that Eq. (9) describes most adequately the regularities of the variation of the retention parameters of *n*-alkanes under conditions of TPGC, and, hence, its coefficients can be used to calculate the retention indices of any substance analyzed under the same conditions.

Calculation of the retention indices of organic compounds using Eq. (9). To determine the retention indices of any organic compound in terms of Eq. (9), we calculated, first of all, coefficients a, b, c, and d by the least-squares method using the adjusted retention times (t') of standard *n*-alkanes and their retention indices $I_{pr} = 100n$. The coefficients of Eq. (9) for seven temperature variation programs during the analysis are listed in Table 2 as examples. It can be seen from these data that the magnitudes of coefficients a and b increase as the rate of temperature variation increases. The effect of the initial temperature of the analysis is especially pronounced for coefficients c and d. The a, b, c, and d values listed in Table 2 make it possible to calculate the retention index of any compound x by substituting its adjusted retention time (t_x'), found under the desired conditions, in Eq. (9). For comparison, the retention indices of homologs of the R_mCOOMe series (m = 1 to 6), calculated using Eqs. (1) and (9), are presented in Table 3. As should be expected (see Fig. 1), the van der Doole and Kratz indices are lower than those found from Eq. (9). In some sections of the chromatogram, the differences between these values amount to ~47 iu. Regarding the reproducibility of the retention indices

Table 2. Coefficients of Eq. (9) for the calculation of retention indices of substances analyzed under various conditions of TPGC

Coefficients	T_0 = 140		T_0 = 160		T_0 = 180		
of Eq. (9)*	r = 2	r = 8	r = 6	r = 8	r = 2	r = 6	r = 8
a	128.2	160.7	136.4	141.7	131.3	132.4	140.6
b	0.151	0.606	0.447	0.603	0.179	0.455	0.621
c	193.4	186.9	214.9	214.3	229.1	230.1	228.9
d	176.1	132.7	206.0	198.7	207.2	205.7	229.0

* Calculated from the data for *n*-alkanes (for T_0 and r, see Table 1).

Table 3. Retention indices for methyl esters of fatty acids, R_mCOOMe, calculated from Eqs. (1) and (9) using various sets (I—III) of reference *n*-alkanes (T_0 = 140 °C, r = 8 °C min^{-1})

m	Equation (9)				Equation (1)			
	I	II	III	*s*	I	II	III	*s*
1	520.5	520.4	520.1	0.6	516.7	512.6	509.5	10.5
2	614.2	614.1	613.2	1.8	611.6	590.1	567.6	64.2
3	709.1	709.0	708.3	1.5	707.2	705.7	660.8	77.1
4	808.9	808.9	808.6	0.6	807.0	786.9	805.6	32.7
5	908.8	908.8	908.9	0.3	907.4	906.1	888.8	30.1
6	1009.3	1009.3	1009.5	0.3	1008.1	991.7	1008.1	27.7

Note: I is C_5—C_{11}; II is C_5, C_7, C_9, and C_{11}; and III is C_5, C_8, C_{10}, and C_{11}.

Table 4. Comparison of the retention indices of methyl esters of fatty acids, R_mCOOMe, calculated from Eq. (9) using a complete set of reference *n*-alkanes (*A*) and using extrapolation technique (*B*) with an incomplete set of *n*-alkanes (T_0 = 140 °C, r = 8 °C min^{-1})

m	Reference *n*-alkanes				
	C_5—C_{14},	C_8—C_{11}		C_5—C_8	
	A	*B*	ΔI_{pr}	*B*	ΔI_{pr}
2	614.6	615.0	−0.4	—	—
3	709.4	708.7	0.7	—	—
4	808.8	—	—	808.5	0.3
5	908.3	—	—	908.5	−0.2
6	1009.0	—	—	1009.9	−0.9
7	1109.7	1110.0	−0.3	1112.7	−3.0
8	1210.7	1209.2	1.5	—	—

calculated for various groups of *n*-alkanes, Eq. (9) is much better than Eq. (1). The standard deviations of the magnitudes of the indices calculated using expression (9) do not exceed 1.8 iu, whereas in the case of Eq. (1), they reach 77.1 iu. The data given in Table 3 indicate that Eq. (9) makes it possible to calculate, within the accuracy of the experiment, the retention indices of compounds using the interpolation technique; this technique does not require a complete set of reference *n*-alkanes. Furthermore, unlike expression (1), the equation that we propose makes it possible to determine the retention indices of compounds by extrapolation (Table 4). It can be seen from Table 4 that the values of I_{pr} for methyl esters of fatty acids can be calculated from Eq. (9) with an accuracy of ±0.9 iu in the absence of the three reference *n*-alkanes, closest to them; when four *n*-alkanes are missing, the accuracy of the calculation is ±3 iu.

Thus, Eq. (9) that we propose allows one to determine the I_{pr} values of compounds when there is no complete set of successive reference *n*-alkanes, using interpolation and extrapolation techniques, and maintaining the accuracy of the calculation.

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References

1. H. van den Doole and P. D. Kratz, *J. Chromatogr.*, 1963, **11**, 463.
2. G. Gastello and P. Parodi, *Chromatographia*, 1971, **4**, 147.
3. D. Messadi, F. Helaimia, S. Ali-Mokhnache, and M. Boumahraz, *Chromatographia*, 1990, **29**, 429.
4. J. Krupcik, P. Cellar, D. Repka, J. Garaj, and G. Guiochon, *J. Chromatogr.*, 1986, **351**, 111.
5. R. V. Golovnya, A. L. Samusenko, and V. A. Lyapin, *Zh. Anal. Khim.*, 1988, **43**, 311 [*Sov. J. Anal. Chem.*, 1988, **43** (Engl. Transl.)].
6. D. N. Grigor'eva, R. V. Golovnya, and A. L. Samusenko, *Izv. Akad. Nauk, Ser. Khim.*, 1994, 642 [*Russ. Chem. Bull.*, 1994, **43**, 591 (Engl. Transl.)].

7. J. C. Giddings, in *Gas Chromatography*, Eds. N. Brenner, J. E. Gallen, and M. D. Weiss, Academic Press, New York, 1962, 57.
8. G. Guiochon, *Anal. Chem.*, 1964, **36**, 661.
9. J. Lee and D. R. Taylor, *Chromatographia*, 1982, **16**, 286.
10. R. V. Golovnya and V. P. Uraletz, *J. Chromatogr.*, 1968, **36**, 276.
11. L. Erdey, J. Takacs, and E. Szalanczy, *J. Chromatogr.*, 1970, **46**, 29.
12. I. G. Zenkevich and B. V. Ioffe, *J. Chromatogr.*, 1988, **439**, 185.
13. T. Wang and Y. Sun, *J. Chromatogr.*, 1987, **390**, 261.
14. R. E. Kaiser and A. J. Rackstraw, *Computer Chromatography*, **1**, Huthig, Heidelberg, 1984,
15. R. V. Golovnya, T. A. Misharina, and V. G. Garbuzov, *Izv. Akad. Nauk SSSR, Ser. Khim.*, 1980, 2503 [*Bull. Acad. Sci. USSR, Div. Chem. Sci.*, 1980, **29**, 1765 (Engl. Transl.)].
16. R. V. Golovnya, D. N. Grigor'eva, and I. L. Zhuravleva, *Izv. Akad. Nauk SSSR, Ser. Khim.*, 1981, 1825 [*Bull. Acad. Sci. USSR, Div. Chem. Sci.*, 1981, **30** (Engl. Transl.)].
17. R. V. Golovnya and N. I. Svetlova, *Chromatographia*, 1988, **25**, 493.

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